THEORETICAL BASES FOR KARYOTYPE EVOLUTION. 1. THE MINIMUM-INTERACTION HYPOTHESIS

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Classical cytogenetics has shown that the nature of the selection acting on a chromosome mutation is a significant factor determining its fixation probability, and hence its likely contribution to karyotype evolution (e.g., John and Lewis 1965; Sybenga 1975). But another important determinant is the rate of occurrence of spontaneous chromosomal mutations, and this rate has been little investigated (except for studies on humans and *Drosophila melanogaster*). On the basis of X-ray irradiation experiments of somatic cells, two theories dealing with this rate have been proposed, the breakage-and-reunion theory (e.g., Sax 1938; Muller 1940) and the exchange theory (Revell 1959, 1974). These theories differ in the mechanisms of chromosomal mutations, but with respect to the so-called two-break rearrangements (i.e., reciprocal translocation and inversion) both theories seem to be stochastically equivalent (Appendix A). Given these circumstances, it is not surprising that most cytologists seem to have accepted without question that the mechanisms of radiation-induced and spontaneous chromosomal mutations do not differ, and therefore the various kinds of chromosomal mutation have the same rates of occurrence for all karyotypes, such that their relative contributions to karyotype evolution are molded principally by purifying selection.

Contrary to the above assumptions, some data indicate that the spontaneous rates of chromosomal mutations deviate markedly from those expected under the exchange theory of Revell (we use here the full term, "random-contact-and-exchange model"). Table 1 gives some data for *Drosophila melanogaster*. For two-break rearrangements induced by X- or γ-rays, the proportion that are reciprocal translocations is either about the same as, or slightly lower than, the value expected under the exchange theory, following expectations determined in Appendix A. This proportion, however, is markedly reduced among spontaneous mutations in laboratory studies or in natural populations. Although the low values found in natural populations might be held to reflect the workings of selection, this interpretation is not tenable for the laboratory studies, because the pre-detection elimination of translocations is expected to be negligible in the experimental

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TABLE 1

RELATIVE FREQUENCIES OF TWO-BREAK REARRANGEMENTS INDUCED BY RADIATION OR OCCURRING SPONTANEOUSLY IN LABORATORY STOCKS OR IN NATURAL POPULATIONS OF *Drosophila melanogaster*

MATERIALS	RECIPROCAL TRANSLOCATION	INVERSION		NUMBER OF REARRANGEMENTS OBSERVED OR REPORTED	SOURCE
		Paracentric	Pericentric		
Theoretically expected	(0.64)	(0.20)	(0.16)	—	Estimate based on the random-contact-and-exchange model; for details, see Appendix A
Induced by	30 (0.63)	14 (0.29)	4 (0.08)	48	Present study
X-rays or γ-rays	8 (0.42)	8 (0.42)	3 (0.16)	19	Woodruff & Ashburner 1978
	34 (0.29)	85 (0.71)		119	Lemke et al. 1978
	756 (0.77)	185 (0.19)	41 (0.04)	982	Lindsley & Grell 1968
Spontaneous	8 (0.14)	48 (0.84)	1 (0.02)	57	Lindsley & Grell 1968
Spontaneous (AW)	1 (0.05)	19 (0.95)	0 (0.00)	20	Yamaguchi & Mukai 1974
Spontaneous (JH)	2 (0.02)	83 (0.91)	6 (0.07)	91	Yamaguchi & Mukai 1974
Spontaneous (C-160)	2 (0.04)	40 (0.70)	15 (0.26)	57	Yamaguchi et al. 1976
Natural population					
Japan	0 (0)	34 (1.00)	0 (0.00)	34	Inoue & Watanabe 1979
USA	0 (0)	55 (0.95)	3 (0.05)	58	Stalker 1976

TABLE 2

RELATIVE FREQUENCIES OF TWO-BREAK REARRANGEMENTS IN NEWBORN BABIES, CLINICAL REFERRALS, AND NATURAL ABORTIONS

Type of Chromosomal Mutation	Theoretically Expected*	Newborn†	Population Survey or Clinical Referral‡	Natural Abortion§
Reciprocal translocation		51	2020	7
	(0.9708)	(0.455)	(0.311)	(0.280)
Centric fusion		53	3126	17
	(0.0008)	(0.473)	(0.481)	(0.680)
Inversion		8	1354	1
	(0.0284)	(0.071)	(0.208)	(0.040)
TOTAL	1.0000	112	6500	25

* Estimate based on the random-contact-and-exchange model. For details, see Appendix A.

† Sergovich et al. 1969; Lubs and Ruddle 1970; Walzer and Gerald 1972; Jacobs et al. 1974; Hamerton et al. 1975; Nielsen and Sillesen 1975.

‡ Borgaonkar et al. 1981. Complex translocation and simple translocation are not cited here, because the former is negligibly low in frequency and the latter seems to be problematic in the definition of terminology.

§ Lauristen et al. 1972; Creasy et al. 1976; Geisler and Kleinebrecht 1978; Kajii et al. 1980; Hassold et al. 1980.

system used by Yamaguchi and his coworkers. Similarly, the relative frequencies of pericentric and paracentric inversions in data from natural populations and from laboratory studies of spontaneous mutations also depart markedly from expectation (table 1).

Data on human chromosome aberrations also suggest a distortion of the spontaneous mutation rates similar to that seen in *Drosophila*. Table 2 shows that in newborns the proportion of chromosomal mutations that are reciprocal translocations is about half that expected, whereas the rate of centric fusion is surprisingly high. A similar trend is evident from population surveys and clinical referrals, and from spontaneous abortions, although the sample size of the latter is too small for this category to be reliable.

A third example is given by ants (fig. 1). As observed by Imai et al. (1977, 1984*a*), reciprocal translocation polymorphisms occur only in species with low chromosome numbers ($n \leq 12$). Now, under the random-contact-and-exchange model, the rate of reciprocal translocations increases with chromosome number, so that these polymorphisms should appear more frequently in the high-numbered species. The trend observed is in the opposite direction. A further unexpected phenomenon is that Robertsonian polymorphisms occur exclusively in high-numbered species ($n > 12$). These data indicate strongly that such chromosome mutations as reciprocal translocation, inversion, and Robertsonian rearrangement have spontaneous rates of occurrence departing markedly from those expected under the random-contact-and-exchange model.

Revell proposed the exchange theory mainly on the basis of data from X-ray irradiation experiments with somatic cells. Spontaneous chromosomal mutations, which contribute to karyotype evolution, should occur in germ cells or in early embryonic cells. Therefore, if the induction mechanisms of radiation-induced and

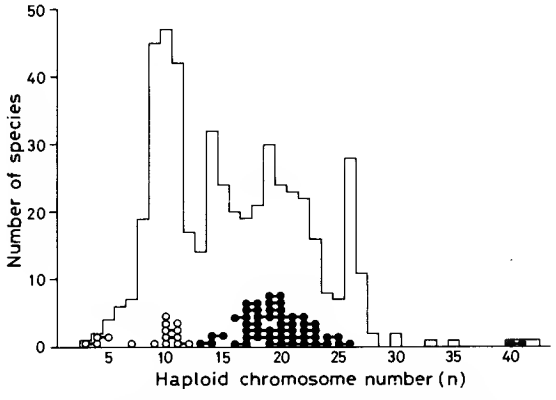

FIG. 1.—Frequency distribution of haploid chromosome numbers (n) in ants, reciprocal translocation polymorphisms (*open circles*), and Robertsonian polymorphisms (*solid circles*). Chromosome numbers involved in the same polymorphic species are connected by short bars. Chromosomal data from Crozier 1975; Crozier 1977; Gōni et al. 1982; Imai and Kubota 1972, 1975; Imai et al. 1977, 1984*a*,*b*. The fraction with $n \leq 12$ is found commonly in hymenopterans other than ants (e.g., sawflies), whereas that with $n > 12$ is characteristic of ants.

spontaneous chromosomal mutations differ substantially, an alternative model might be an improvement on the random-contact-and-exchange model. We discuss here such an alternative model and its implications for karyotype evolution.

MODELS AND DATA

Possible Mechanisms of Spontaneous Chromosomal Mutations

Ever since Muller (1938, 1940) and Sax (1938), such structural chromosomal mutations as reciprocal translocations, inversions, and centric fusions have been interpreted, under the breakage-and-reunion theory, as simple two-break rearrangements leading to the occurrence probabilities given in Appendix A. We have noted above that the exchange theory is stochastically equivalent to the breakage-and-reunion theory except for the proposed mechanism for the induction of chromosomal mutations (see also Appendix A). A further guiding factor, however, is that those spontaneous chromosomal mutations that contribute to karyotype evolution must occur either in the germ cells or in early embryonic cells. The actual mechanisms leading to DNA strand breaks or exchanges in germ cells or early embryonic cells must be considered when discussing the occurrence probabilities of spontaneous chromosomal mutations.

Radiation, chemical mutagens, mobile elements (transposons), sister-chromatid exchanges (SCE's), mis-resolution of chromosome interlockings, and crossovers are demonstrated or theoretically available mechanisms leading to DNA strand breaks or exchange. Among these, radiation and chemical mutagens may be less likely to contribute to spontaneous chromosomal mutation because, as will be discussed later, the spectra of chromosomal mutations seem to differ for radiation-induced and spontaneous rearrangements (e.g., see table 1 and also fig. 10).

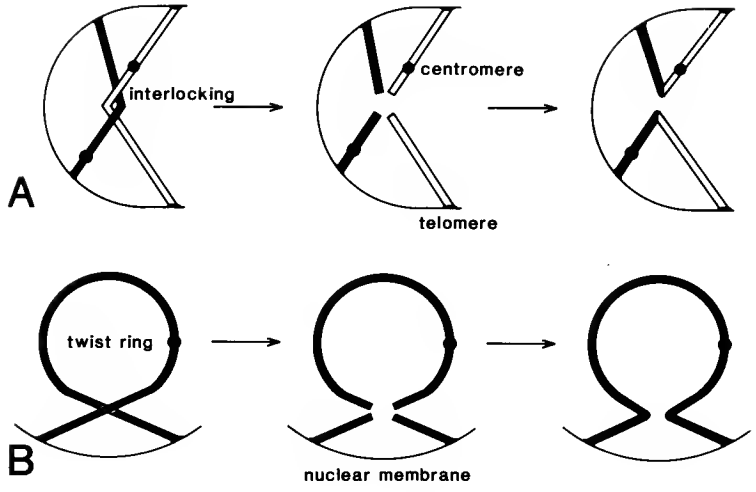

FIG. 2.—Chromosome mutations induced by a mis-resolution of interlockings: *A*, reciprocal translocation; *B*, inversion. For details, see text.

Transposons may play a significant role in *Drosophila* (Shapiro 1983) and are believed to cause spontaneous chromosomal mutations in *Drosophila melanogaster* (table 1; Yamaguchi and Mukai 1974; Mukai, pers. comm.), but further data are needed to establish the importance of this factor in eukaryotes generally. Spontaneous SCE (John and Miklos 1979; Tsuji 1982) may also be an important factor promoting centric fusion if it occurs in premeiotic gonial cells in association with a heterochromatin aggregation, though spontaneous SCE's do not occur in some organisms (e.g., *D. melanogaster*).

We consider the mis-resolution of interlockings and crossovers probably the most important mechanisms to be discussed here, because these phenomena are universally observed in eukaryotes (Rasmussen and Holm 1980; Holm and Rasmussen 1984; Wettstein et al. 1984) and they occur at much higher frequencies (one to about 60 per cell) than the others (e.g., less than 0.1 per cell for SCE; Tsuji 1982). Errors in the resolution of interlockings at late zygotene or early pachytene seem to make up the most important factor inducing reciprocal translocation and inversion (fig. 2). Theoretically, *all* spontaneous chromosomal mutations contributing to karyotype evolution can result from a crossover between nonhomologous chromosomes or at the intersecting point of a twisted bivalent (fig. 3). (Since these crossover types were not familiar in classical cytogenetics, we use the term "hetero-site crossover.") The formation of a nonhomologous synaptonemal complex in *Bombyx mori* (Wettstein et al. 1984) offers significant support for this view.

Regularity in Chromosome Configuration in Pachytene Nuclei

Some cytogeneticists have known for years that interphase nuclei have a specific chromosome arrangement in somatic cells, referring to it by various

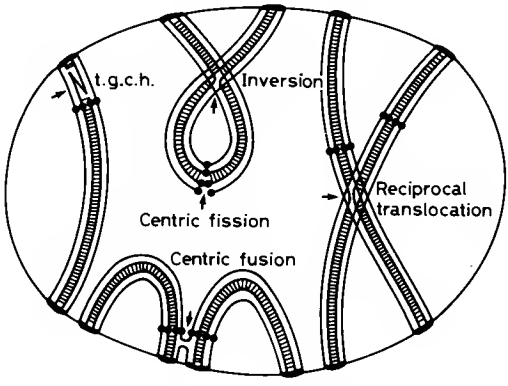

FIG. 3.—Schematic representation of the suspension-arch structure of pachytene chromosomes and chromosomal mutations that are theoretically expected when the crossover mechanism "hetero-site crossover" is used. Note that the terminals of each bivalent are fixed on the nuclear membrane by the so-called telomere, resulting in a configuration universal in eukaryotes, which we call the "suspension-arch structure." Short bars denote synaptonemal complex (SC). The unit chromosome (sister chromatid or monad) comprising each tetrad or bivalent is represented by a solid line. t.g.c.h., tandem growth of constitutive heterochromatin in short arms. Arrows indicate the sites of chromosomal mutations.

names: Rabl configuration (Rabl 1885; Cremer et al. 1982); polarized chromosome arrangement (Fox 1966); telomere-telomere attachment (Sved 1966; Ashley and Pocock 1981); or satellite association (Nakagome 1969; Hemel 1971). This nonrandom configuration of chromosomes significantly affects the occurrence probability of chromosomal mutations. As mentioned above, however, chromosomal mutations that contribute to karyotype evolution must occur either in the germ cells or in early embryonic cells. In connection with this, we also noted that the mis-resolution of interlockings, or hetero-site crossover, may be the most important mechanism for spontaneous chromosomal mutations. We therefore now discuss the nonrandom chromosome configuration at early meiotic prophase, especially at pachytene.

Recent advances in electron microscopy have shown that chromosomes are laterally associated with their homologues by the synaptonemal complex during zygotene and pachytene (e.g., Moses 1968; Wettstein et al. 1984). The resulting bivalent is associated with the nuclear membrane at both termini by being fixed to it by the basal knob (Solari 1970) or by the terminal-attachment plaque (Moses 1977) or telomere. These findings indicate that the arrangement of bivalents in meiotic prophase nuclei is highly structured and nonrandom. Because of its shape, we term the resulting chromosome configuration at pachytene the "suspension-arch structure" (fig. 3). Reconstructions of synaptonemal complex from electron micrographs indicate that the suspension-arch structure is probably universal in eukaryotes, for example, in the fungi *Neurospora crassa* (Gillies 1979) and *Sordaria macrospora* (Zickler 1977), maize (Gillies 1973), the silkworm *Bombyx mori* (Rasmussen 1976), and humans (Holm and Rasmussen 1977), as well as several other cases (Wettstein et al. 1984). It is therefore likely that the relative occur-

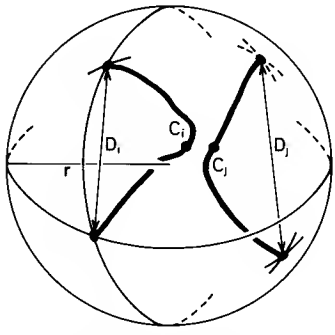

FIG. 4.—A schematic representation for estimating the effect of the suspension-arch structure affecting the occurrence probability of reciprocal translocation. r, radius; C_i and C_j, chromosomes i and j, respectively. D_i and D_j, distances between the two terminals in chromosomes i and j. The nucleus is schematically represented by a sphere with radius r containing two chromosomes, i and j, which were selected arbitrarily from a given haploid karyotype and which have their terminals fixed randomly onto the nuclear membrane. Chromosomes i and j can move freely, except for their terminals. We assume that if they contact (interact) they exchange their arms at the point of contact (i.e., reciprocal translocation). Thus, the total frequency of interaction of chromosomes i and j estimated with all the combination of chromosome numbers ($C_{i=1,\ldots,n}$, $C_{j=1,\ldots,n}$, $i \neq j$) corresponds to the probability of the occurrence of reciprocal translocation per cell.

rence rates of spontaneous chromosomal mutations will be affected by the suspension-arch structure. The so-called bouquet configuration may also affect the occurrence rate of chromosomal mutations, especially tandem fusions or centric fusions (for details, see Appendix B).

Simulation of the Occurrence Probabilities of Reciprocal Translocations and Inversions

Reciprocal translocations.—Let the nucleus of primary spermatocytes or primary oocytes be a sphere of radius r (fig. 4). Let i and j be two randomly chosen chromosomes from a given karyotype, K, with haploid number n, and let C_i and C_j be their respective sizes in terms of a percentage of the combined length of all the chromosomes in a haploid set. Let the two ends of each chromosome be placed randomly on the inside surface of the sphere, which is the interphase nucleus. Now we can ask whether the two chromosomes are close enough to come into physical contact. We follow here the exchange theory of Revell: if they are in contact, a chromosomal exchange leading to a translocation can occur, but an exchange is impossible if they do not touch. Of course, the actual probability of an exchange also depends on the lengths of the two chromosomes, but for now we will ignore this additional factor and only ask how often two chromosomes will be in sufficiently close proximity to allow an exchange. This principle for the simulation can be extended to all n chromosomes of the haploid set, enabling us to estimate the relative abundance of the number of chromosome pairs in close proximity in a nucleus. This abundance ratio is naturally a function of the nuclear radius, r, and the haploid chromosome number, n. Using five different species

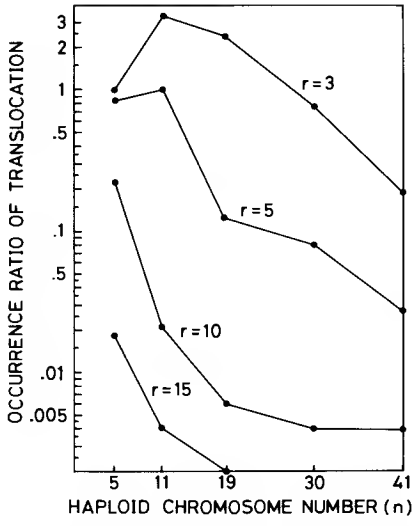

FIG. 5.—Effects of nuclear volume and chromosome numbers influencing the probability of the occurrence of reciprocal translocation. *r*, radius of nucleus (arbitrary value; see fig. 4). The estimates were made using the karyotypes of Australian bulldog ants (*Myrmecia*) having $n = 5$, 11, 19, 30, and 41 (for details, see Imai et al. 1977). The frequency of reciprocal translocation is represented as a proportion of the value for the case $r = 3$ and $n = 5$. Reciprocal translocations decrease with increasing nuclear volume or chromosome numbers.

having $n = 5$, 11, 19, 20, and 41, we simulated the relative abundance for various values of r (3, 5, 10, 15; fig. 5). Each point plotted represents an average derived from 2000 simulations.

The five karyotypes used in these simulations are taken from species of the Australian bulldog ant *Myrmecia*: *M. pilosula* ($n = 5$), *M. nigrocincta* (11), *M. gulosa* (19), *M. fulvipes* (30), and *M. pyriformis* (41) (for details of their karyotypes, see Imai et al. 1977). Because there is no information on nuclear size for these species, we used a range of values, letting r equal 3, 5, 10, and 15. Inspection of the simulation results brings out two features: when nuclear size (r) is fixed, the frequency of reciprocal translocation decreases as the chromosome number increases; and when chromosome number is fixed, this frequency decreases as nuclear size increases.

There is one exception to these trends, and that is in the curve of $r = 3$, in which there is an initial rise before a fall. This exception may be an artifact of our estimation strategy, in that we considered only the numbers of bivalents in proximity but not the lengths of chromosome involved. Because the average chromosome length is larger in low-numbered than in high-numbered karyotypes, more than two reciprocal translocations are expected in the former but not in the latter. Our estimation strategy is therefore likely to underestimate the reciprocal translocation frequency in the case of $r = 3$ and $n = 5$.

Inversion.—Let the length of the short arm, long arm, and whole chromosome for bivalent i be S_i, L_i, and $C_i = S_i + L_i$, respectively; all lengths are given as

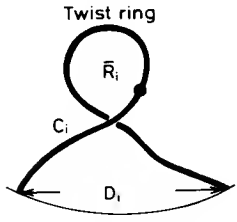

FIG. 6.—A schematic representation for inversion formation by the suspension-arch-structure model (see also figs. 2, 3). For simplifying the estimation, chromosome i (C_i) at pachytene is represented as monad (sister chromatid). D_i, the distance between two terminals fixed on the nuclear membrane. The chromosome can move freely, except for the terminals, and form a special ring configuration (twist ring; $\bar{R}_i$). If a crossover occurs at the intersecting point of the twist ring, the rearrangement expected is an inversion.

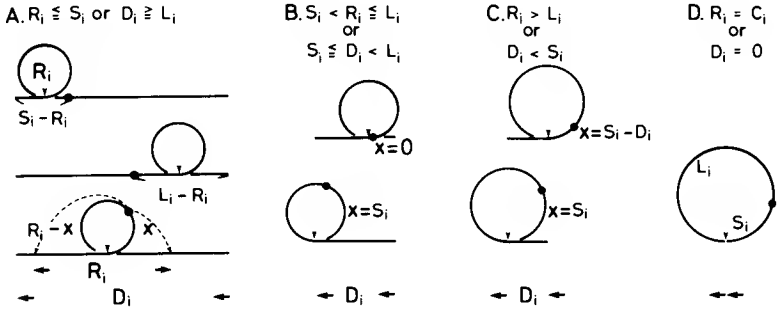

FIG. 7.—A schematic representation for estimating the occurrence probabilities of paracentric inversion (In_{para}) and pericentric inversion (In_{peri}) by the suspension-arch-structure model. D_i, distance between two terminals of chromosome i fixed on the nuclear membrane. R_i, the maximum twist ring. S_i and L_i, the short arm and long arm, respectively, of chromosome i (C_i), where $C_i = S_i + L_i$ and $R_i = C_i - D_i$. x and $R_i - x$, the short arm and long arm, respectively, in the maximum twist ring. Arrows mark the intersecting point of each twist ring. For details, see Appendix B.

percentages of the combined lengths of the haploid chromosome set. Both arms are attached to the nuclear membrane, yielding the suspension-arch structure (fig. 3). The distance, D_i, between the two terminals (telomeres) is $0 \leq D_i \leq C_i$ (fig. 6). The random movements of a bivalent under this structural restriction are expected to yield frequently a special chromosome configuration, resulting from the bivalent twisting and forming a ring, the "twist ring" (fig. 6). If a crossover or error in the resolution of an interlocking occurs involving the intersection point of a twist ring, the result will be an inversion (cf. figs. 2, 3, and 6). If the twist ring is wholly within one arm, a paracentric inversion results (fig. 7*A*, top and center), but if the twist ring includes the centromere, a pericentric inversion results (fig. 7*A*, bottom). This model parallels that of Revell's exchange hypothesis (1959, 1974) explaining chromatid exchanges induced by radiation.

We can now estimate the relative occurrence rate of paracentric inversions

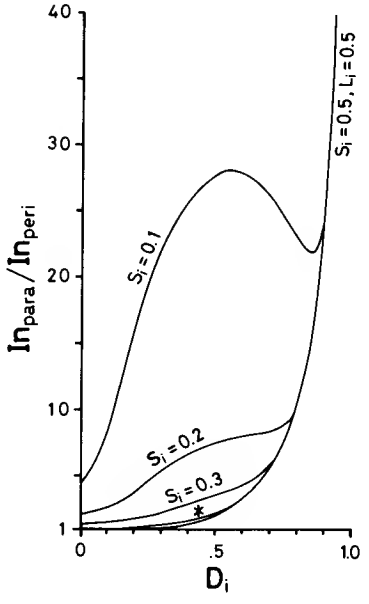

FIG. 8.—Effects of chromosome shapes (M, SM, ST, or A) and terminal distance (D_i) affecting the occurrence probability of paracentric inversion (In_{para}) and pericentric inversion (In_{peri}). S_i and L_i, the short arm and long arm of chromosome i with $C_i = 1$. *Asterisk*, case $S_i = 0.4$ (M). Case $D_i = 0$ corresponds to the estimation by the random-contact-and-exchange model (see Appendix A). The frequency of paracentric inversions relative to that of pericentric inversions increases with D_i and decreases with S_i.

(In_{para}) versus pericentric inversions (In_{peri}). The rate can be given as the ratio $In_{para/peri}$, and the effects on it of inter-telomere distance, D_i, for metacentrics (S_i = 0.4 and 0.5), submetacentrics ($S_i = 0.3$), subtelocentrics ($S_i = 0.2$), and acrocentrics ($S_i = 0.1$), with $C_i = 1$. The details of this estimation are given in Appendix B; the results are summarized in figure 8. Under this model, paracentric inversions are increasingly the more common type as D_i increases for all centromere positions. Metacentric chromosomes form a particularly important case, because the much-studied chromosome 2 of *Drosophila melanogaster* is metacentric. Our results, if correct, indicate that the observed predominance of paracentric inversions in *D. melanogaster* ($In_{para/peri} = 6.7–8.6$; table 1) is readily interpretable by the suspension-arch-structure model assuming $D_i = 0.72–0.76$. No preferential elimination of pericentric inversions by selection need be invoked.

DISCUSSION

The Minimum-Interaction Hypothesis

We propose that a major factor channeling karyotype evolution may have been selection for reduced opportunity for spontaneous chromosomal mutations. This selection arises because spontaneous chromosomal mutations, especially recip-

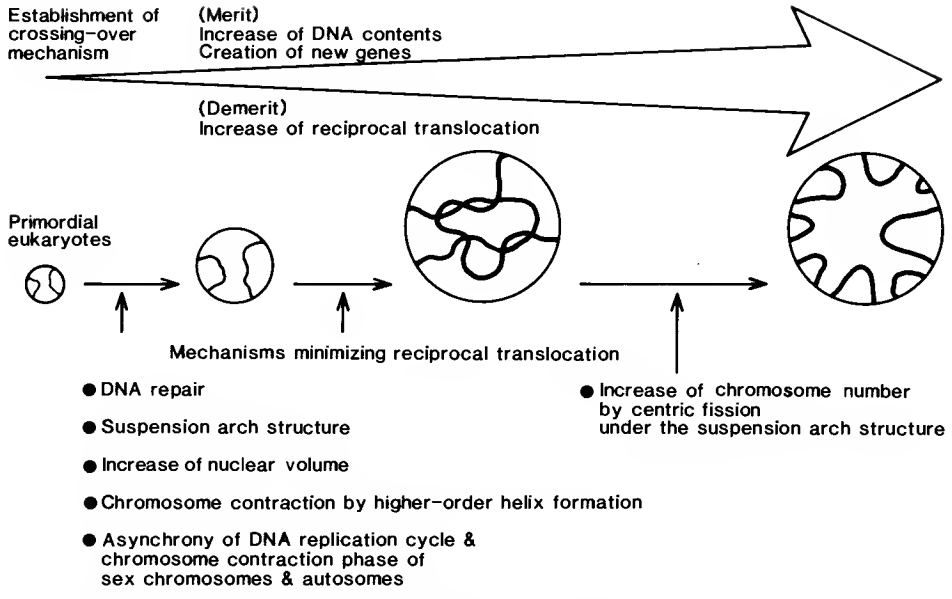

FIG. 9.—Schematic representation of the minimum-interaction hypothesis. For details, see text.

rocal translocations, tend to reduce the fitness of heterozygotes. Because spontaneous chromosomal mutations require the physical proximity of chromosomes, we term this hypothesis the "minimum-interaction hypothesis." We will now discuss the reasoning that leads us to this hypothesis.

There has been a general tendency in eukaryotes for an increase of nuclear DNA, and hence of genome size, with the attainment of higher grades of organization (Lewin 1980; Cavalier-Smith 1985). Such increases can occur by polyploidy, but also by unequal crossover leading to intrachromosomal gene duplication (Ohno 1970). When it has involved gene duplication, the increase of genome size has probably been instrumental in allowing various groups the opportunity for evolutionary advance, but it also increases the rate of the production of deleterious spontaneous chromosomal mutations.

In eukaryotes selection seems to have acted through at least five avenues to reduce the genetic risks imposed by spontaneous chromosomal mutation (fig. 9): (1) improved DNA repair, (2) suspension-arch structure, (3) increase of nuclear volume in oocytes and spermatocytes, (4) increased chromosome contraction by higher-order helix formation of the constituent chromatin, and (5) asynchrony of the DNA replication cycle and chromatin-contraction phases of sex chromosomes and autosomes. The first mechanism is well known to cytogeneticists, but the remainder, especially 2 and 3, seem not to be as familiar in the present context. The suspension-arch structure may have evolved as a risk-avoidance mechanism by disorganizing the bouquet configuration under selection to minimize the rate of spontaneous chromosomal mutation. The bouquet configuration would have

played a major cellular function of which we are so far ignorant, especially in the early step of eukaryote evolution. As suggested in Appendix B, however, the frequency of reciprocal translocation and centric fusion is increased greatly by the bouquet configuration. Note that in the absence of the suspension-arch structure, the rate of reciprocal translocation increases with chromosome number (Appendix A), whereas when nuclear organization includes the suspension-arch structure, the reciprocal translocation rate decreases with increasing chromosome number (fig. 5, Appendix B).

Whatever the evolutionary factor leading to the formation of the suspension-arch structure, it further affects the probabilities of the occurrence of spontaneous chromosomal mutations. One effect is to reduce these probabilities as nuclear volume increases (fig. 9). Indeed, there is a general tendency for the nuclear volume to increase with increasing genome size (Cavalier-Smith 1985). Chromosome number also affects these probabilities, to an extent that increases as the ratio of genome size to nuclear volume increases. The reason for this trend is straightforward. For the same genome size, the mean chromosome size will be larger in low-numbered karyotypes than in high-numbered ones. Therefore, given the suspension-arch structure, interaction between nonhomologous chromosomes (which results in reciprocal translocations) will be fewer in high-numbered than in low-numbered karyotypes (fig. 9).

The remarkable contraction of chromatin fibers by higher-order helix formation reported by many authors (e.g., Leth Bak and Zeuthen 1978; Sedat and Manuelidis 1978; Worcel 1978) may also have evolved because of selection for reduced risk of spontaneous chromosomal mutation. Without such a contraction, the effective length of chromosomes, especially at pachytene, is much greater, thereby increasing the probability of spontaneous chromosomal mutations.

Quite a few cases of X-autosome translocations (yielding multiple sex-chromosome systems) are known for animals (White 1973; Austin and Edwards 1981), but species having these systems are distinctly in the minority. We suggest that, without the asynchrony of the DNA-replication and chromatin-contraction phases, multiple sex-chromosome systems would be overwhelmingly the rule rather than the exception. Incorporation of loci (other than the sex-determining ones, of course) into the sex-chromosome system may be undesirable because of the general (but not universal) need for dosage compensation for such loci (Bull 1983).

A final point about the minimum-interaction hypothesis concerns the marked difference between the spectrum of spontaneous chromosomal mutations and that of radiation-induced changes (table 1). We make two suggestions to resolve this anomaly. (1) The great majority of spontaneous chromosomal mutations occur in synaptonemal complexes via crossovers and the mis-resolution of interlockings. (2) Radiation and some chemical mutagens (minor causes of spontaneous chromosomal mutation), by contrast, can act at any stage when chromatids (homologous or not) are in contact (fig. 10), and can lead to a characteristic spectrum of chromosomal mutations, which can be described more profitably by Revell's exchange theory than by the breakage-and-reunion theory of Sax or Muller. These proposals are susceptible to test.

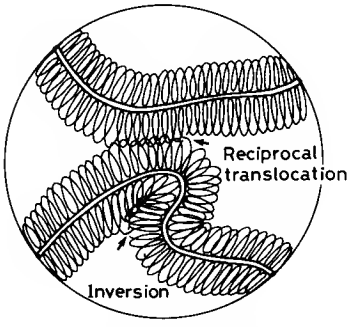

FIG. 10.—A possible resolution for the paradox that reciprocal translocation and inversions are induced abundantly by radiation, whereas they are rarely spontaneous in *Drosophila melanogaster* (table 1). We assume that spontaneous chromosomal mutations are induced mainly by the crossover mechanism or a mis-resolution of interlockings. If this assumption is correct, then chromosomal mutations depend on the interaction of synaptonemal complexes between bivalents, mediated by the suspension-arch structure. Bivalents at pachytene contact each other at the chromatin level, but they do not interact unless the DNA strands are broken or damaged by radiation or chemicals.

Modes of Karyotype Evolution under the Minimum-Interaction Hypothesis

According to the minimum-interaction hypothesis, the pattern of karyotype evolution will be determined largely by the ratio of genome size to nuclear volume of oocytes or spermatocytes at pachytene (fig. 9). If this ratio is low, reciprocal translocations occur rarely (relative to cases with high ratios). In such instances, increases in chromosome number will not be strongly favored, and low-numbered karyotypes may be preserved for long stretches. *Drosophila melanogaster* may be a case in point (table 1). The well-known evidence that amphibians have rather low chromosome numbers in spite of their high DNA contents may also be explained by the large size of their oocyte nuclei. There is further evidence that nuclear volume increases with increasing genome size in eukaryotes (Cavalier-Smith 1985), suggesting that the increase in nuclear volume may have played an important role for reducing the genetic risks of chromosomal mutations in many eukaryotes. In contrast, if the ratio of genome size to nuclear volume is high, selection will be strong for an increase in chromosome number to provide a compensating reduction in the risk of reciprocal translocations. The nonrandom distribution of reciprocal translocation and Robertsonian polymorphisms found in ants (fig. 1) may be an example of this phenomenon.

In no genetic system, of course, can we rule out the occasional reduction of chromosome numbers through centric fusion; such cases represent "back eddies" to the mainstream of karyotype evolution (Imai and Crozier 1980). Consider the case of $r = 10$ in figure 5. Here, the incidence of reciprocal translocations is reduced only very slightly ($0.006 \rightarrow 0.004$) by a great increase in chromosome number ($n = 19 \rightarrow 41$). For such a genetic system, an increase in chromosome number is not favored strongly by selection for risk avoidance, and fusions may be readily fixed. In this connection, high rates of fusion for particular chromosomes

(as for human D- and G-group chromosomes, in which fusion is thought to result from satellite association; Nakagome 1969, 1973; Hemel 1971) may lead to low-numbered karyotypes to a degree set by the ratio of genome size to nuclear volume. Such a reduction has not been fixed in humans, but can be inferred to have occurred in some groups otherwise characterized by high-numbered karyotypes (e.g., *Mus musculus*, Gropp et al. 1972, 1982; *Dipodomys*, Stock 1974; and *Rattus rattus*, Yosida 1980). But the evidence is that chromosome numbers in any eukaryote groups have tended to increase by centric fission, rather than decrease by centric fusion (Imai 1978; Imai and Crozier 1980). The minimum-interaction hypothesis provides a theoretical framework for understanding such trends.

SUMMARY

Data from the literature (tables 1, 2; fig. 1) show that the relative probabilities of occurrence for the various categories of spontaneous chromosomal mutations do not match those predicted by both the random-contact-and-exchange model (basically equivalent to the exchange theory of Revell) and the random-breakage-and-reunion model (i.e., the breakage-and-reunion theory of Sax or of Muller). These models do not take into account recent findings that, during the meiotic prophase and especially at pachytene, eukaryote chromosomes are attached by each end to the nuclear membrane, leading to a configuration we term the "suspension-arch structure" (fig. 3).

We recalculated the relative probabilities of the occurrence of spontaneous chromosomal mutations given the suspension-arch structure and assuming that these rearrangements arise from errors in the resolution of interlockings between bivalents and a special type of crossover that we call the "hetero-site" crossover (figs. 2, 3, 6, 7). From these calculations we found that the relative probability of the occurrence of reciprocal translocations (the most fitness-damaging rearrangement) declines with increases in chromosome number and in nuclear volume (fig. 5). We also found that paracentric inversions occur increasingly more often than pericentric ones as the centromere position becomes more terminal and the distance between attachments to the nuclear membrane becomes greater (this distance increases as nuclear volume increases; fig. 8). These results are in accord with the cytogenetic data from *Drosophila*, humans, and ants.

A puzzling phenomenon is that the relative rates of radiation-induced chromosomal mutations differ greatly from those calculated under the suspension-arch-structure model and from rates of spontaneous chromosomal mutations. We propose the testable hypotheses that (1) most spontaneous chromosomal mutations occur in synaptonemal complexes and involve crossovers and errors in the resolution of interlockings, and (2) radiation and chemical mutagens allow rearrangements when chromosomes intersect at any stage (fig. 10).

These considerations lead to the "minimum-interaction hypothesis," which states that karyotype evolution has been in large part shaped by selection to reduce the occurrence of such fitness-reducing spontaneous chromosomal mutations as reciprocal translocations. Some of the response to this selection is the result of the improvement of DNA-repair mechanisms, increased contraction of

the chromosomes caused by higher-order helix formation, and the development of sex-chromosome heteropycnosis. We concentrate on examining two further, interacting responses in the light of the hypothesis. One of these is increase in nuclear volume (fig. 9), but if the ratio of genome size to nuclear volume is high, then an increase in chromosome number, caused by such factors as centric fission, is adaptive because it reduces the occurrence of reciprocal translocation. Although chromosome number can be reduced by centric fusion, such instances seem to be "back eddies" in the mainstream of karyotype evolution.

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APPENDIX A

The Probabilities of the Occurrence of Translocation, Inversion, and Centric Fusion under the Random-Contact-and-Exchange Model

Let C_i be the size of chromosome i, and S_i and L_i, respectively, be the short and long arms of the chromosome, where $C_i = S_i + L_i$ and their sizes are represented by their relative length against the total chromosome length of a given haploid karyotype (unit genome). We assume that two chromosomes (bivalents) selected arbitrarily from a given karyotype contact at an arbitrary point on the chromosomes, and a complete exchange occurs at the intersection (fig. A1). Such an exchange induces a translocation. If each chromosome twists to form a ring at an arbitrary point (twist ring; see fig. 6 and Appendix B) and a complete exchange occurs at the intersection, we can expect an inversion. These estimations are mainly based on Revell's exchange theory, but to simplify estimations we assume random contact among chromosomes. Hence, we use the term "random-contact-and-exchange model."

The probability of the occurrence of inversion (*In*) and translocation (*Tr*) per haploid karyotype is proportional to the probability of two randomly occurring breakpoints occurring on the same chromosome, or to the probability of these points falling on nonhomologues, and is given by

$$In = \sum_{i=1}^{n} C_i^2 \tag{A1}$$

and

$$Tr = \sum_{i=1}^{n} \sum_{j=1}^{n} C_i C_j = 1 - \sum_{i=1}^{n} C_i^2, \tag{A2}$$

where C_i and C_j are two nonhomologous chromosomes ($i \neq j$).

In a hypothetical karyotype having n equal-sized chromosomes, $C_i = 1/n$ and then $In = n\,(1/n)^2 = 1/n$, so that *In* decreases as n increases. This means that inversion occurs less frequently in high-numbered karyotypes than in low-numbered ones. From equation (A2), we obtain $Tr = 1 - 1/n$, so that the value of *Tr* increases with n. This indicates that

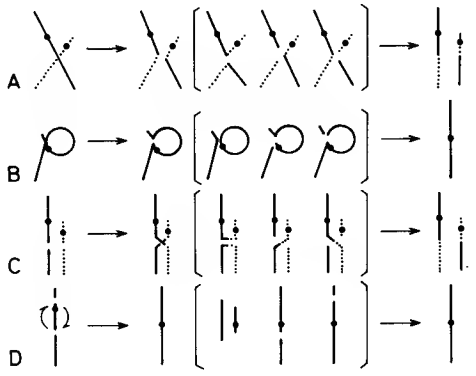

FIG. A1.—A schematic representation of chromosomal mutations by the random-contact-and-exchange model (*A*, *B*) and the random-breakage-and-reunion model (*C*, *D*). *A and C*, reciprocal translocation (interchange). *C and D*, pericentric inversion (intrachange). Incomplete types are shown in parentheses. To simplify estimations, we assumed exchange or reunion of only complete types. Such simplification does not introduce any bias to the estimations if the ratios of the complete type to the incomplete types are always constant in both rearrangement types and if we consider the relative probability of occurrence between translocation and inversion. Note that the probabilities of occurrence for both rearrangements are given by the same formulas in both models (see eqs. A1 and A2), indicating that both models are stochastically equivalent even though they differ in the mechanisms postulated.

translocation occurs more frequently in high-numbered karyotypes than in low-numbered ones.

The occurrence probabilities of paracentric (In_{para}) and pericentric inversion (In_{peri}) are

$$In_{para} = \sum_{i=1}^{n} S_i^2 + \sum_{i=1}^{n} L_i^2 \tag{A3}$$

and

$$In_{peri} = 2 \sum_{i=1}^{n} S_i L_i . \tag{A4}$$

In_{para} and In_{peri} change with the shape of chromosomes. In metacentrics in the strict sense (M)($S_i = L_i = C_i/2$), $In_{para} = In_{peri} = 2\,\Sigma_{i=1}^{n} (C_i/2)^2$, whereas $In_{para} > In_{peri}$ in submetacentrics (SM), subtelocentrics (ST), and acrocentrics (A).

The probability of centric fusion (Fu) is

$$Fu = \sum_{i=1}^{n'} \sum_{j=1}^{n'} S_i' S_j' , \tag{A5}$$

where n' is the number of acrocentrics and S_i' and S_j' are short arms of two nonhomologous acrocentrics ($i \neq j$). Fu increases with the number of acrocentrics (n'), but the value is remarkably small, because $S_i' \leq 0.006$ and $S_j' \leq 0.006$ (i.e., less than 0.6% of the total length of the haploid chromosome set) by the definition of acrocentrics in mammals (Imai 1976, 1978).

The same argument is also true under the breakage-and-reunion theory, if two breaks occur randomly on two different chromosomes (translocation) or on the same chromosome

(inversion), and if a complete-type reunion occurs between the naked chromosome ends. Because we emphasize randomness, we use the term the "random-breakage-and-reunion model" in this text. Note that for chromosomes to touch at two randomly selected points and then for an exchange to occur at the points is stochastically equivalent to arbitrarily selecting two points on the chromosome(s) and then having two breakages and two complete-type reunions occurring at the points (fig. A1), though the biological mechanisms involved are different. The breakage-and-reunion theory and the exchange theory were both proposed on the bases of data from radiation experiments with somatic cells, where translocation and inversion are recognized as two-hit rearrangements in the breakage-and-reunion theory but they are induced by a single ionizing track in the exchange theory (Abrahamson and Wolf 1976). The minimum-interaction hypothesis proposed here agrees with the exchange theory. We suggest, however, that radiation-induced and spontaneous chromosomal mutations differ substantially in their mechanisms, and that hetero-site crossovers (fig. 3) and mis-resolution of interlockings (fig. 2) may be the two major mechanisms for spontaneous chromosomal mutations.

APPENDIX B

The Probabilities of the Occurrence of Pericentric and Paracentric Inversions under the Suspension-Arch-Structure Model

Let $\bar{R}_i$ and R_i be the size of the average twist ring ($\bar{R}_i \leq C_i - D_i$; fig. 6) and the maximum (possible) twist ring ($R_i = C_i - D_i$; fig. 7), where C_i is the size of chromosome i and D_i is the distance between telomeres. If two breaks occur outside the maximum twist ring, no inversion is expected. However, all the paired breaks that occur in the maximum twist ring have the potential to induce inversions. The estimates are made with the following four categories, which are classified in terms of R_i or D_i: (A) $R_i \leq S_i$ or $D_i \geq L_i$ (fig. 7*A*); (B) $S_i < R_i \leq L_i$ or $S_i \leq D_i < L_i$ (fig. 7*B*); (C) $R_i > L_i$ or $D_i < S_i$ (fig. 7*C*); and (D) $R_i = C_i$ or $D_i = 0$ (fig. 7*D*).

Category A is subdivided into three cases: (1) the maximum twist ring is involved only in the short arm (fig. 7*A*, top); (2) it lies only in the long arm (fig. 7*A*, center); or (3) the ring includes the centromere (fig. 7*A*, bottom). The effective arm sizes in which the maximum twist ring can appear for each case are, respectively, $(S_i - R_i)$, $(L_i - R_i)$, and R_i. The probability of the occurrence of an inversion expected in the maximum twist ring is R_i^2. In cases 1 and 2, only paracentric inversion occurs, and the probabilities expected for the short arm, $In_{para}(S)$, and for the long arm, $In_{para}(L)$, are

$$In_{para}(S) = R_i^2(S_i - R_i)/D_i \tag{B1}$$

and

$$In_{para}(L) = R_i^2(L_i - R_i)/D_i . \tag{B2}$$

Both paracentric and pericentric inversions occur in case 3. Let the sizes of the short arm and the long arm in the maximum twist ring be x and $R_i - x$ (fig. 7*A*, bottom), respectively, where $0 \leq x \leq R_i$. The probability of paracentric inversion in this case, $In_{para}(C)$, is

$$\begin{aligned} In_{para}(C) &= \frac{R_i}{D_i}(R_i^2)\frac{\int_0^{R_i}\left[x^2 + (R_i - x)^2\right]dx}{\int_0^{R_i}\left[x^2 + (R_i - x)^2\right]dx + \int_0^{R_i} 2x(R_i - x)dx} \\ &= 2R_i^3/3D_i . \end{aligned} \tag{B3}$$

Now, the total probability of the occurrence of paracentric inversion (In_{para}) is

$$In_{para} = In_{para}(S) + In_{para}(L) + In_{para}(C) = R_i^2[C_i - (4/3)R_i]/D_i . \tag{B4}$$

Similarly, the probability of pericentric inversion (In_{peri}) is

$$In_{peri} = (R_i/D_i)\, R_i^2 \int_0^{R_i} 2x(R_i - x)dx = R_i/3D_i. \tag{B5}$$

From equations (B4) and (B5), the relative ratio of In_{para} to In_{peri} is

$$In_{para}/In_{peri} = (3C_i - 4R_i)/R_i = (4D_i - C_i)/(C_i - D_i). \tag{B6}$$

Equations (B4) and (B5) are basically applicable to the estimation of In_{para} and In_{peri} of categories B and C, though we need some modifications, as follows: $In_{para}(S) = 0$ and $0 \le x \le S_i$ in category B, and $In_{para}(S) = In_{para}(L) = 0$ and $S_i - D_i \le x \le S_i$ in category C. Now, In_{para}/In_{peri} for category B is

$$\begin{aligned} In_{para}/In_{peri} &= \frac{(L_i - R_i)R_i^2 + \int_0^{S_i}\left[x^2 + (R_i - x)^2\right]dx}{\int_0^{S_i} 2x(R_i - x)dx} \\ &= [3D_i(C_i - D_i)^2 - I_1]/I_1, \end{aligned} \tag{B7}$$

where $I_1 = 3(C_i - D_i)S_i^2 - 2S_i^3$. For category C,

$$\begin{aligned} In_{para}/In_{peri} &= \frac{\int_{S_i - D_i}^{S_i}\left[x^2 + (R_i - x)^2\right]dx}{\int_{S_i - D_i}^{S_i} 2x(R_i - x)dx} \\ &= [3D_i(C_i - D_i)^2 - I_2]/I_2, \end{aligned} \tag{B8}$$

where $I_2 = 3D_i(C_i - D_i)(2S_i - D_i) - 2D_i(3S_i^2 - 3S_iD_i + D_i^2)$.

Category D shown in figure 7*D* is an extreme case, in which $D_i = 0$ and $R_i = C_i$, both terminals of chromosome i are attached to form a ring on the nuclear membrane. Since the entire short and long arms are included in the maximum twist ring ($R_i = C_i = S_i + L_i$), we can expect inversion (i.e., twist ring) corresponding to all the paired breaks occurring in the short and/or long arms. In_{para}, In_{peri}, and $In_{para/peri}$ are therefore

$$In_{para} = S_i^2 + L_i^2, \tag{B9}$$

$$In_{peri} = 2S_iL_i, \tag{B10}$$

and

$$In_{para/peri} = (S_i^2 + L_i^2)/2S_iL_i. \tag{B11}$$

Note that equations (B9) and (B10) are essentially identical to equations (A3) and (A4) if $n = 1$. This means that the random-contact-and-exchange model is a special case of the exchange model given by the suspension-arch structure. The case of $D_i = 0$ is actually realized in the so-called bouquet configuration. In the bouquet configuration, terminals of all bivalents distribute in a narrow area on the nuclear membrane, and thus bivalents can contact each other almost randomly. If so, the probabilities of the occurrence of reciprocal translocation and inversion will be given by the formulas (A1) and (A2), and tandem fusion and centric fusion will increase significantly. Since the bouquet configuration is observed in some protozoans (Raikov 1982), such a structure would have played some significant role in the early steps of eukaryote evolution. In higher organisms having high DNA contents and small nuclear volumes, disorganization of the bouquet configuration may be more evolutionarily adaptive, as reflected in mammals.

LITERATURE CITED

Abrahamson, S., and S. Wolff. 1976. Re-analysis of radiation-induced specific locus mutations in the mouse. Nature (Lond.) 264:715–719.

Ashley, T., and N. Pocock. 1981. A proposed model of chromosomal organization in nuclei at fertilization. Genetica (The Hague) 55:161–169.

Austin, C. R., and R. G. Edwards. 1981. Mechanisms of sex differentiation in animals and man. Academic Press, London.

Borgaonkar, D. S., R. Shaffer, W. C. Reed, L. G. Jackson, T. Casey, K. Levitsky, and I. Gonzalez. 1981. Repository of chromosomal variants and anomalies in man. 8th Listing. Wilmington Medical Center, Wilmington, Del.

Bull, J. J. 1983. Evolution of sex-determining mechanisms. Benjamin/Cummings, Menlo Park, Calif.

Cavalier-Smith, T. 1985. Cell volume and the evolution of eukaryotic genome size. Pages 105–184 *in* T. Cavalier-Smith, ed. The evolution of genome size. Wiley, Chichester, England.

Creasy, M. R., J. A. Crolla, and E. D. Alberman. 1976. A cytogenetic study of human spontaneous abortions using banding techniques. Hum. Genet. 31:177–196.

Cremer, T., C. Cremer, H. Baumann, E.-K. Luedtke, K. Sperling, V. Teuber, and C. Zorn. 1982. Rable's model of the interphase chromosome arrangement tested in Chinese hamster cells by premature chromosome condensation and laser-UV-microbeam experiments. Hum. Genet. 60:46–56.

Crozier, R. H. 1975. Hymenoptera: animal cytogenetics. Vol. 3. Insecta 7. Gebruder Borntrager, Berlin.

———. 1977. Genetic differentiation between populations of the ant *Aphaenogaster "rudis"* in the southern United States. Genetica (The Hague) 47:17–36.

Fox, D. P. 1966. The effects of X-rays on the chromosomes of locust embryos. II. Chromatid interchanges and the organization of the interphase nucleus. Chromosoma (Berl.) 20: 173–194.

Geisler, M., and J. Kleinebrecht. 1978. Cytogenetic and histologic analyses of spontaneous abortions. Hum. Genet. 45:239–251.

Gillies, C. B. 1973. Ultrastructural analysis of maize pachytene karyotypes by three-dimensional reconstruction of the synaptonemal complexes. Chromosoma (Berl.) 43:145–176.

———. 1979. The relationship between synaptonemal complexes, recombination nodules and crossing over in *Neurospora crassa* bivalents and translocation quadrivalents. Genetics 91:1017.

Gōni, B., H. T. Imai, M. Kubota, M. Kondo, H. S. Yong, and Y. P. Tho. 1982. Chromosome observations of tropical ants in western Malaysia and Singapore. Annu. Rep. Natl. Inst. Genet. (Jpn.) 32:71–73.

Gropp, A., H. Winking, L. Zech, and H. Muller. 1972. Robertsonian chromosomal variation and identification of metacentric chromosomes in feral mice. Chromosoma (Berl.) 39:265–288.

Gropp, A., H. Winking, and C. Redi. 1982. Consequences of Robertsonian heterozygosity: segregational impairment of fertility versus male-limited sterility. Pages 115–134 *in* P. G. Crosignani, B. L. Rubin, and M. Fraccaro, eds. Genetic control of gamete production and function. Academic Press, London.

Hamerton, J. L., N. Canning, M. Ray, and S. Smith. 1975. A cytogenetic survey of 14,069 newborn infants. 1. Incidence of chromosome abnormalities. Clin. Genet. 8:223–243.

Hassold, T., N. Chen, J. Funkhouser, T. Jooss, B. Manuel, J. Matsuura, A. Matsuyama, C. Wilson, J. A. Yamane, and P. A. Jacobs. 1980. A cytogenetic study of 1000 spontaneous abortions. Ann. Hum. Genet. 44:151–178.

Hemel, J. O., van. 1971. Studies on human chromosome satellite association. Bronder-Offset N. V., Rotterdam.

Holm, P. B., and S. W. Rasmussen. 1977. Three-dimensional reconstructions of meiotic chromosomes in human spermatocytes. Chromosomes Today 6:83–93.

———. 1984. The synaptonemal complex in chromosome pairing and disjunction. Chromosomes Today 8:104–116.

Imai, H. T. 1976. Further evidence and biological significance for non-random localization of the centromere on mammalian chromosomes. J. Theor. Biol. 61:195–203.

———. 1978. On the origin of telocentric chromosomes in mammals. J. Theor. Biol. 78:619–637.
Imai, H. T., and R. H. Crozier. 1980. Quantitative analysis of directionality in mammalian karyotype evolution. Am. Nat. 116:537–569.
Imai, H. T., and K. Kubota. 1972. Karyological studies of Japanese ants (Hymenoptera, Formicidae) III. Chromosoma (Berl.) 37:193–200.
———. 1975. Chromosome polymorphism in the ant, *Pheidole nodus*. Chromosoma (Berl.) 51: 391–399.
Imai, H. T., R. H. Crozier, and R. W. Taylor. 1977. Karyotype evolution in Australian ants. Chromosoma (Berl.) 59:341–393.
Imai, H. T., C. Baroni-Urbani, M. Kubota, G. P. Sharma, M. N. Narashimhanna, B. C. Das. A. K. Sharma, A. Sharma, G. B. Deodikar, V. G. Vaidya, and M. R. Rajasekarasetty. 1984*a*. Karyological survey of Indian ants. Jpn. J. Genet. 59:1–32.
Imai, H. T., W. L. Brown, Jr., M. Kubota, H. S. Yong, and Y. P. Tho. 1984*b*. Chromosome observations of tropical ants in western Malaysia (2). Annu. Rep. Natl. Inst. Genet. (Jpn.) 34:66–69.
Inoue, Y., and T. K. Watanabe. 1979. Inversion polymorphisms in Japanese natural populations of *Drosophila melanogaster*. Jpn. J. Genet. 54:69–82.
Jacobs, P. A., M. Melville, S. Ratcliffe, A. J. Keay, and J. Syme. 1974. A cytogenetic survey of 11,680 newborn infants. Ann. Hum. Genet. 37:359–376.
John, B., and K. R. Lewis. 1965. The meiotic system: protoplasmatologia. Springer-Verlag, Vienna.
John, B., and G. L. G. Miklos. 1979. Functional aspects of satellite DNA and heterochromatin. Int. Rev. Cytol. 58:1–114.
Kajii, T., A. Ferrier, N. Niikawa, H. Takahara, K. Ohma, and S. Avirachan. 1980. Anatomic and chromosomal anomalies in 639 spontaneous abortuses. Hum. Genet. 55:87–98.
Lauristen, J. G., J. Jonasson, A. J. Therkelsen, F. Lass, J. Lindsten, and G. B. Peterson. 1972. Studies on spontaneous abortions: fluorescence analysis of abnormal karyotypes. Hereditas 71:160–163.
Lemke, D. E., J. Tonzetich, and M. V. Shumeyko. 1978. Resistance to radiation-induced chromosomal rearrangements in *Drosophila simulans*. *Drosophila* Inf. Serv. 53:159–161.
Leth Bak, A., and J. Zeuthen. 1978. Higher-order structure of mitotic chromosomes. Cold Spring Harbor Symp. Quant. Biol. 42:367–377.
Lewin, B. 1980. Gene expression. 2. Eukaryotic chromosomes. 2d ed. Wiley, New York.
Lindsley, D. L., and E. H. Grell. 1968. Genetic variations of *Drosophila melanogaster*. Carnegie Inst. Wash., Publ. 627.
Lubs, H. A., and F. H. Ruddle. 1970. Chromosomal abnormalities in the human population: estimation of rates based on New Haven newborn study. Science (Wash., D.C.) 169:495–497.
Moses, M. J. 1968. Synaptonemal complex. Annu. Rev. Genet. 2:363–412.
———. 1977. Synaptonemal complex karyotyping in spermatocytes of the Chinese hamster (*Cricetulus griseus*). Chromosoma (Berl.) 60:99–125.
Muller, H. J. 1938. The remaking of chromosomes. Collect Net. (Woods Hole) 8:182–195, 198. (Reprinted *in* Studies in genetics: the selected papers of H. J. Muller. Indiana University Press, Bloomington, 1962.)
———. 1940. Bearings of the *Drosophila* work on systematics. Pages 185–268 *in* J. Huxley, ed. The new systematics. Clarendon, Oxford.
Nakagome, Y. 1969. DNA replication studies of human D-group chromosomes in satellite associations. Cytogenetics (Basel) 8:296–303.
———. 1973. G-group chromosomes in satellite associations. Cytogenet. Cell Genet. 12:336–341.
Nielsen, J., and I. Sillesen. 1975. Incidence of chromosome aberrations among 11,148 newborn children. Humangenetik 30:1–12.
Ohno, S. 1970. Evolution by gene duplication. Springer-Verlag, New York.
Rabl, C. 1885. Über zellteilung. Morphol. Jahrb. 10:214–330.
Raikov, I. B. 1982. The protozoan nucleus: morphology and evolution. Cell Biol. Monogr. 9.
Rasmussen, S. W. 1976. The meiotic prophase in *Bombyx mori* females analyzed by three-dimensional reconstructions of synaptonemal complex. Chromosoma (Berl.) 54:245–293.
Rasmussen, S. W., and P. B. Holm. 1980. Mechanisms of meiosis. Hereditas 93:187–216.

Revell, S. H. 1959. The accurate estimation of chromatid breakage, and its relevance to a new interpretation of chromatid aberrations induced by ionizing radiations. Proc. R. Soc. Lond. B, Biol. Sci. 150:563–589.

———. 1974. The breakage-and-reunion theory and the exchange theory for chromosomal aberrations induced by ionizing radiations: a short history. Adv. Radiat. Biol. 4:367–416.

Sax, K. 1938. Chromosome aberrations induced by X-rays. Genetics 23:494–516.

Sedat, J., and L. Manuelidis. 1978. A direct approach to the structure of eukaryotic chromosomes. Cold Spring Harbor Symp. Quant. Biol. 42:331–350.

Sergovich, F., G. H. Valentine, A. T. L. Chen, R. A. H. Kinch, and M. S. Smout. 1969. Chromosome aberrations in 2,159 consecutive newborn babies. N. Engl. J. Med. 280:851–855.

Shapiro, J. A. 1983. Mobile genetic elements. Academic Press, New York.

Solari, A. J. 1970. The spatial relationship of the X and Y chromosomes during meiotic prophase in mouse spermatocytes. Chromosoma (Berl.) 29:217–236.

Stalker, H. D. 1976. Chromosome studies in wild populations of *D. melanogaster*. Genetics 82: 323–347.

Stock, A. D. 1974. Chromosome evolution in the genus *Dipodomys* and its taxonomic and phylogenetic implications. J. Mammal. 55:505–526.

Sved, J. A. 1966. Telomere attachment of chromosomes: some general and cytological consequences. Genetics 53:747–756.

Sybenga, J. 1975. Meiotic configuration. Springer-Verlag, Berlin.

Tsuji, H. 1982. Presence of a base line level of sister chromatid exchange in somatic cells of *Drosophila melanogaster* in vivo and in vitro. Genetics 100:259–278.

Walzer, S., and P. S. Gerald. 1972. Chromosome abnormalities in 11,154 newborn infants. Am. J. Hum. Genet. 24:38.

Wettstein, D. von, S. W. Rasmussen, and P. B. Holm. 1984. The synaptonemal complex in genetic segregation. Annu. Rev. Genet. 18:331–413.

White, M. J. D. 1954, 1973. Animal cytology and evolution. 2d, 3d eds. Cambridge University Press, London.

Woodruff, R. C., and M. Ashburner. 1978. The frequency of X-ray-induced chromosome breakage in the sibling species *Drosophila melanogaster* and *Drosophila simulans*. Am. Nat. 112: 456–459.

Worcel, A. 1978. Molecular architecture of the chromatin fiber. Cold Spring Harbor Symp. Quant. Biol. 42:313–324.

Yamaguchi, O., and T. Mukai. 1974. Variation of spontaneous occurrence rates of chromosomal aberrations in the second chromosomes of *Drosophila melanogaster*. Genetics 78:1209–1221.

Yamaguchi, O., R. A. Cardellino, and T. Mukai. 1976. High rates of occurrence of spontaneous chromosome aberrations in *Drosophila melanogaster*. Genetics 83:409–422.

Yosida, T. H. 1980. Cytogenetics of the black rat: karyotype evolution and species differentiation. University of Tokyo Press, Tokyo.

Zickler, D. 1977. Development of the synaptonemal complex and the "recombination nodules" during meiotic prophase in the seven bivalents of the fungus *Sordaria macrospora* Auersw. Chromosoma (Berl.) 61:289–316.